Self-assembled complexes of biopolymers and charged membranes

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Summary

Relation between electrostatic self-assembly
and biomineralization

- ‘Molecular casting’ using DNA-cationic membrane complexes

  Crystallographic control of ‘biomineralized’ CdS nanorods

- Control of pore size, shape, & chemistry in biopolymer-membrane systems

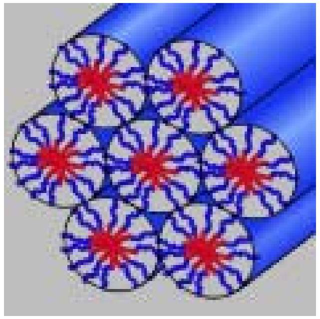
  ** ‘Design rules’ of large-pore lamellar complexes: virus-membrane assembly*
  ** Fourier reconstruction of ion locations within virus-membrane complex*

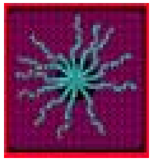
- Like-charge attraction and assembly between anionic membranes and anionic polyelectrolytes mediated by divalent ions

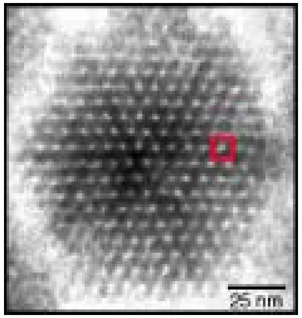
  Structural polymorphism and implications for controlled release

How to crystallize inorganic crystals in soft matter systems?

Solution templating: size and shape control



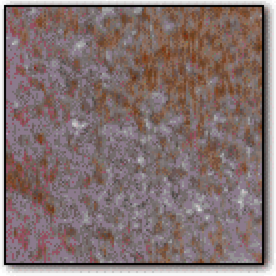




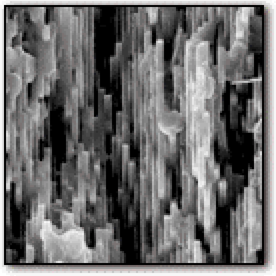
see for example: Kresge et al., 1992; Braun et al., *1996;* Brinker, 1998

Biomineralization: hierarchical structure & crystallographic orientation

abalone shell



Outer layer: Calcite



Inner layer: microlaminate composite of calcium carbonate crystals (aragonite) and proteins with a fracture-toughness 3,000-times greater than crystal alone

see for example: Addadi & Weiner, 1992; *Biomineralization,* S. Mann, 2002

Why can't we simply copy Nature?

- It is known that anionic proteins are involved in guiding biomineralization, although many of the relevant proteins have yet not been identified

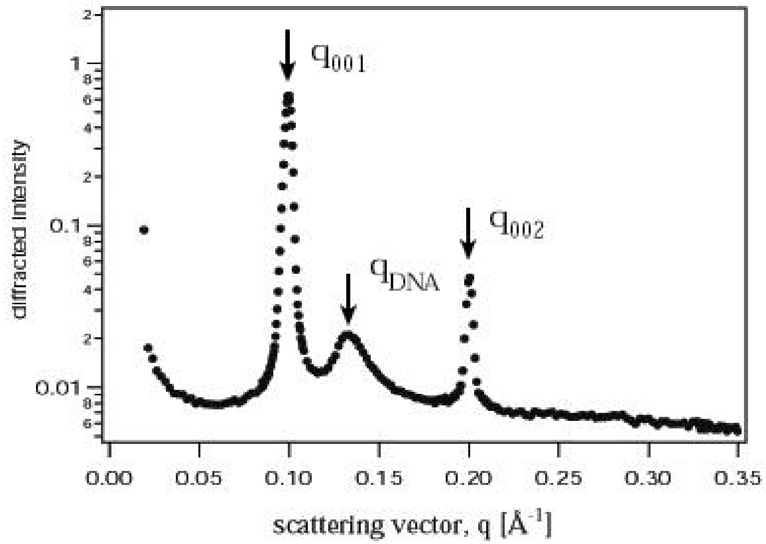
- Not all the roles for these proteins are known

- Proteins difficult to purify, result in small quantities

→ ***Study simplified system: examine mineralization behavior of biomolecular templates with structures, charge distributions, phase behavior, and self-assembly characteristics that can be controlled precisely.***

DNA-cationic membrane complexes

Raedler, Koltover, Salditt, Safinya **Science** (1997)



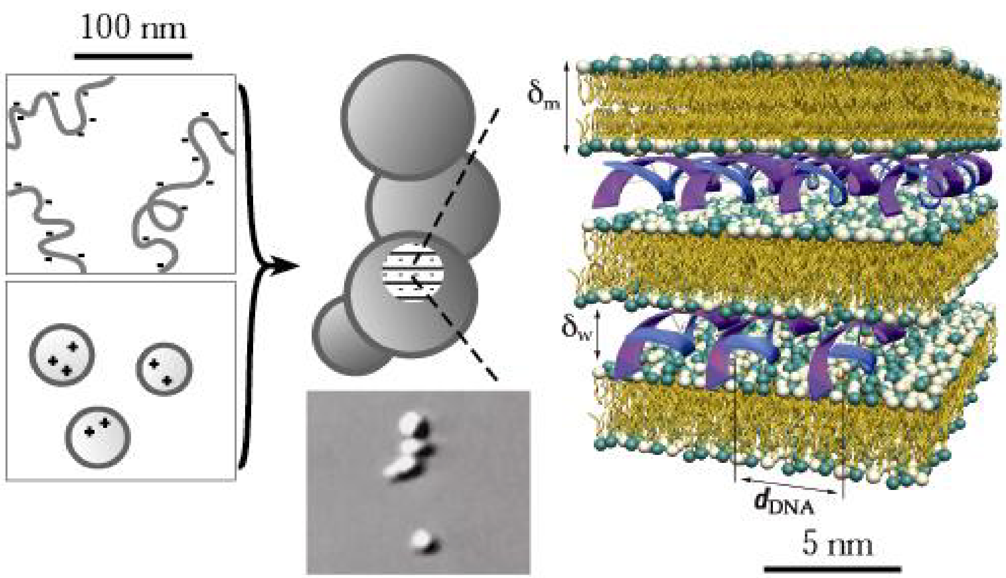
$q_{001} = 2\pi/d$

$q_{DNA} = 2\pi/d_{DNA}$

$d = 66Å = \delta_m + \delta_w$

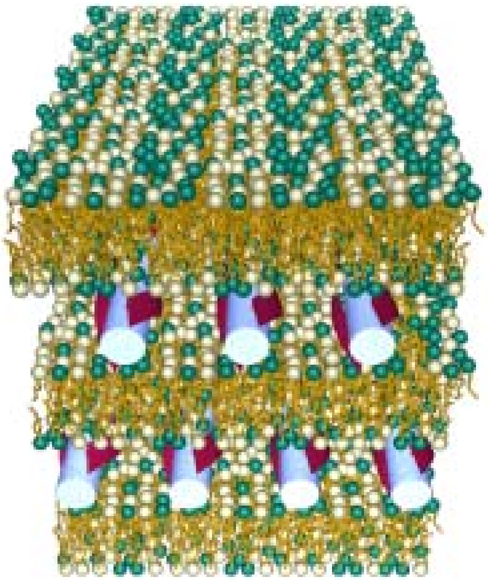
$\delta_m = 42Å$

- Synthetic DNA-cationic membrane complexes: originally conceived for non-viral gene therapy
- 1-D lattice of DNA chains intercalated between lipid bilayer sheets
- Inter-DNA spacing can be controlled by membrane charge density (2.5nm-6.5nm)
- Nanoporous system with tunable pore size



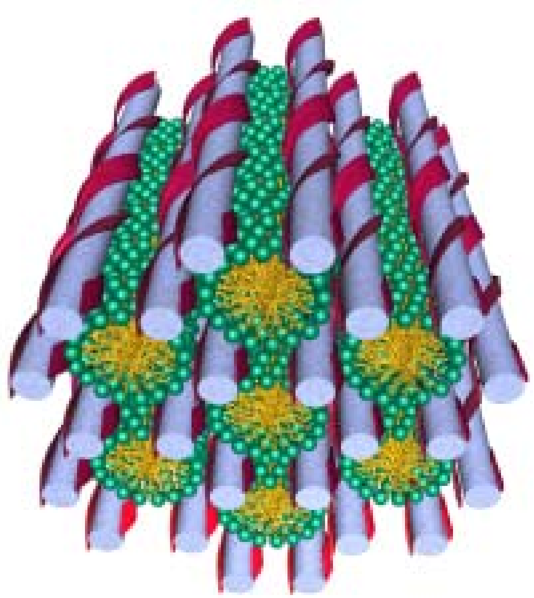
Highly compact, self-assembled structure

Polymorphism of DNA-lipid phases



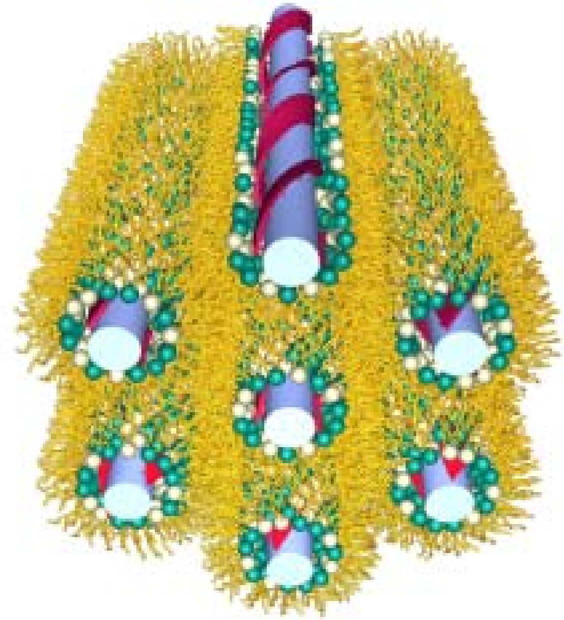
Lamellar (L_{α})

Radler et al., (1997)



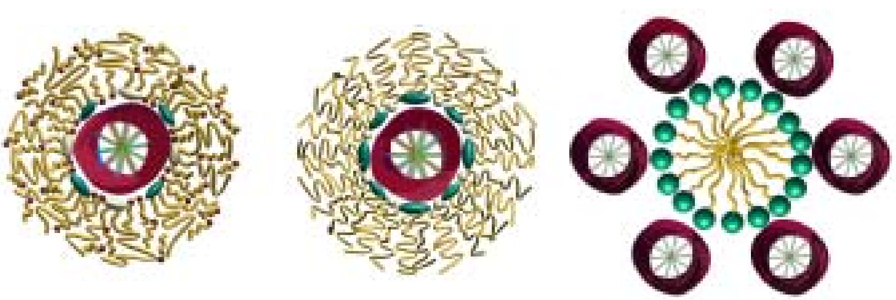
Hexagonal (H_{I})

Krishnaswamy et al., (2003)



Inverted hexagonal (H_{II})

Koltover et al., (1998)



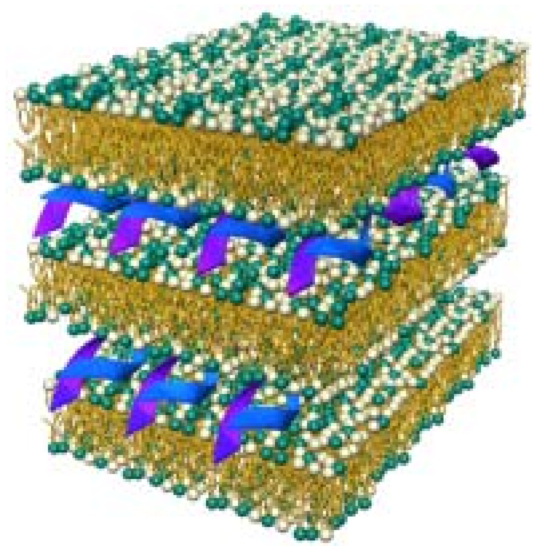
- Tune membrane bending modulus (ex. cosurfactants, etc.)
- Change shape of amphiphilic molecule

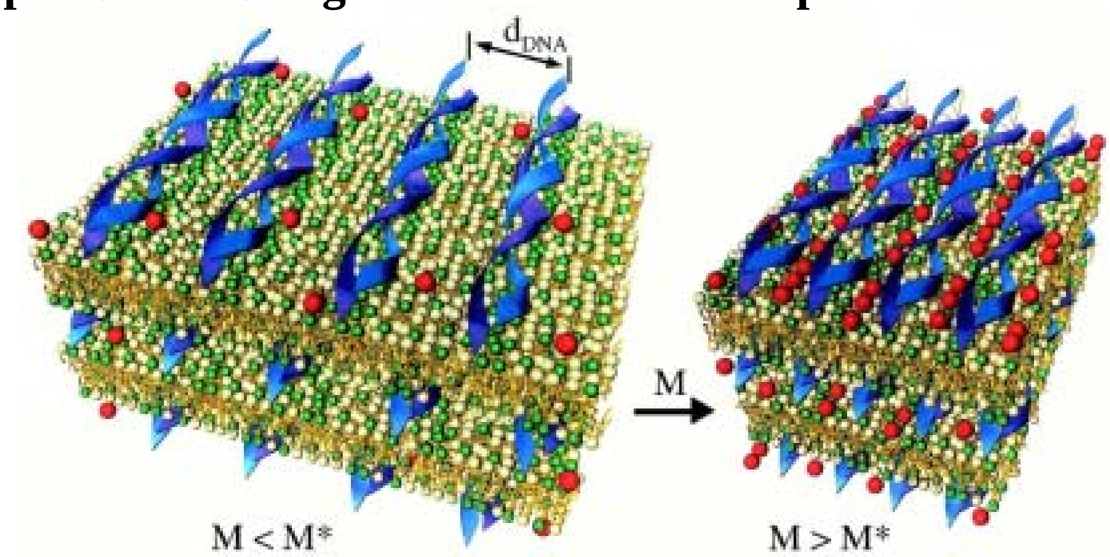
Can self-assembled DNA-cationic membrane templates be used to ‘direct’ crystallization?

- Control not just pore size and shape, but chemical structure and charge distribution on pore walls
- Complementary to amphiphile-based templating

How do DNA-membrane complexes interact with ions?

Organization of ion precursors using DNA-membrane complexes





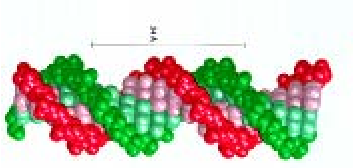
A DNA-membrane complex originally conceived for gene therapy, with tunable pore sizes

2-D condensation of DNA rods by divalent counterions → confined arrays of metal ions inside nanoporous matrix

Pictures adapted from Rädler J.O. et al, Science (1997) and Koltover I. et al, PNAS (2000).

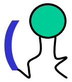
Components for cationic membrane-DNA complexes

Anionic biopolymer

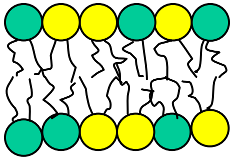


λ-phage DNA, calf thymus DNA

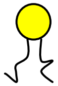
2e$^-$ / 3.4Å

Cationic lipid ()

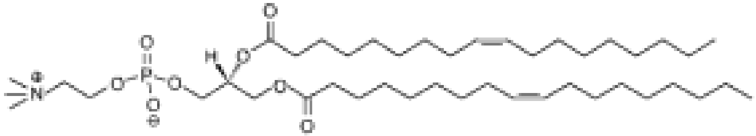
DOTAP



1 positive charge / 70Å^2

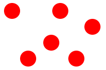
Neutral lipid ()

DOPC



Cadmium ion Cd^{2+}

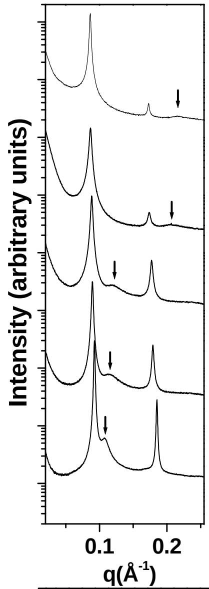
(precursor to CdS)



Organization of Cd ions in DNA-membrane complex

SAXS: structure of DNA-membrane-ion complex

ICPS: density of condensed ions in complex

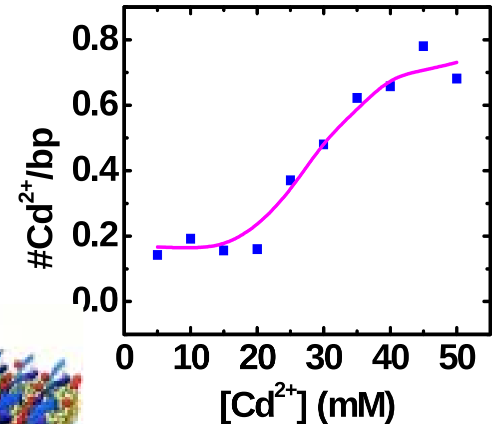


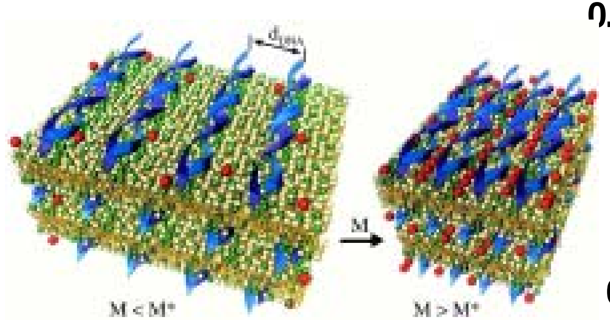
$[Cd^{2+}]$	d_{DNA}
40 mM	28.8Å
30 mM	30.5Å
20 mM	52.3Å
10 mM	54.6Å
0 mM	58.1Å

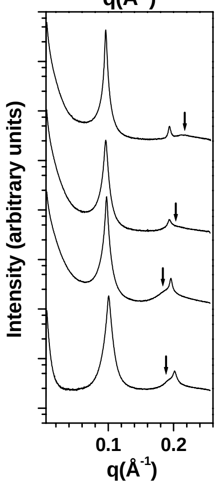
isoelectric

30/70 DOTAP/DOPC

Low membrane charge density





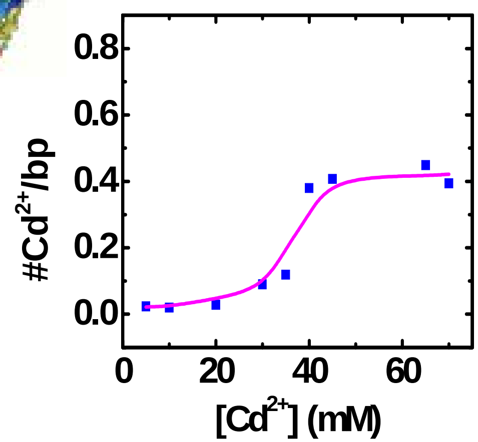


$[Cd^{2+}]$	d_{DNA}
40 mM	28.9Å
30 mM	30.6Å
20 mM	32.9Å
0 mM	32.9Å

isoelectric

70/30 DOTAP/DOPC

High membrane charge density

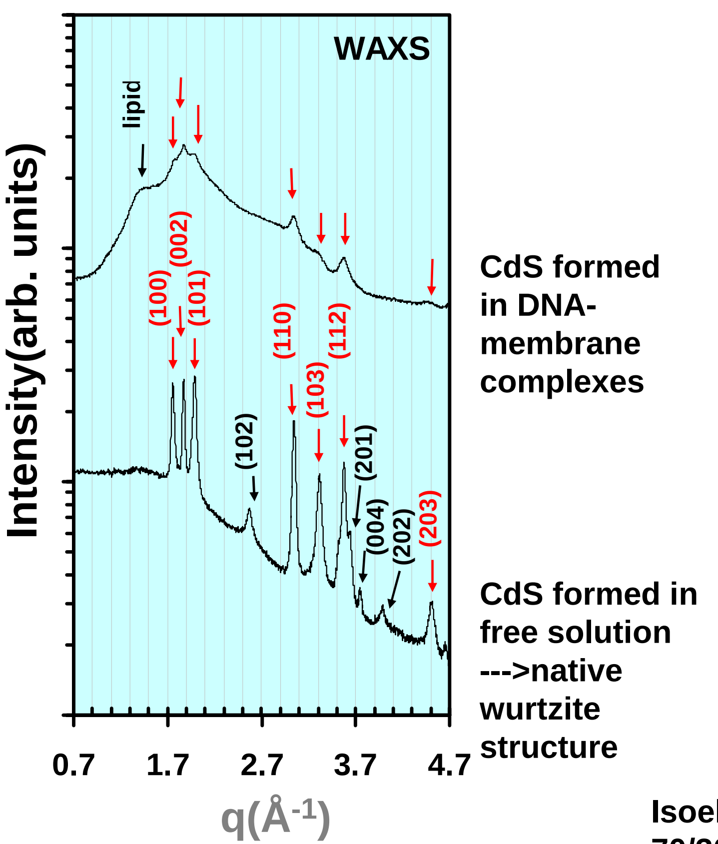


Control of inter-ion spacing: High membrane charge density
→ fewer Cd^{2+} ions organized into 2-D DNA-membrane array

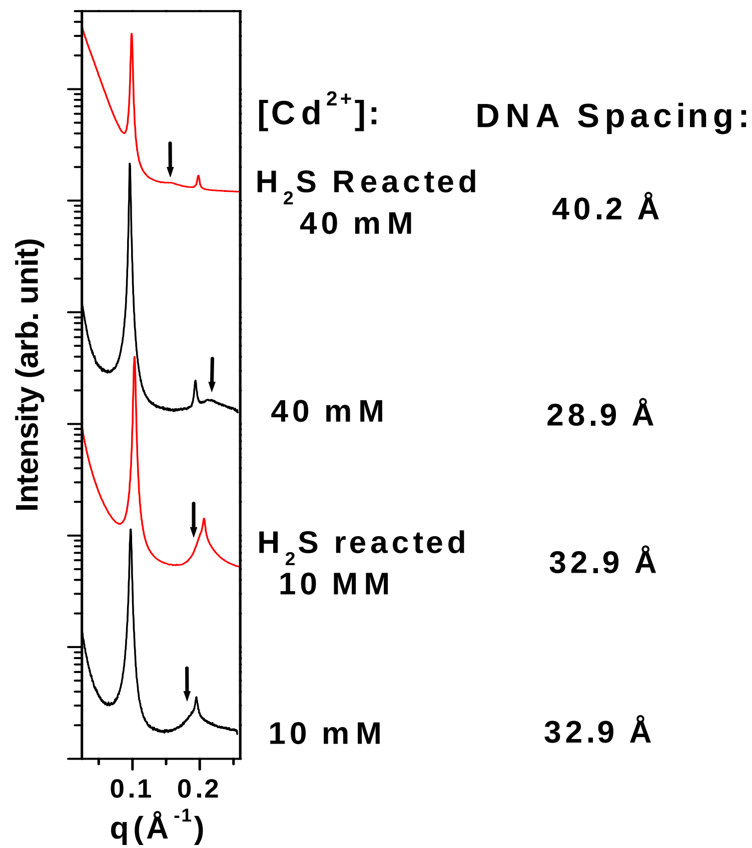
Structural evolution of DNA-membrane template during CdS growth

Cd ions confined in DNA-membrane complexes react with H_2S to form CdS nanorods with wurtzite structure

WAXS: structure of templated CdS

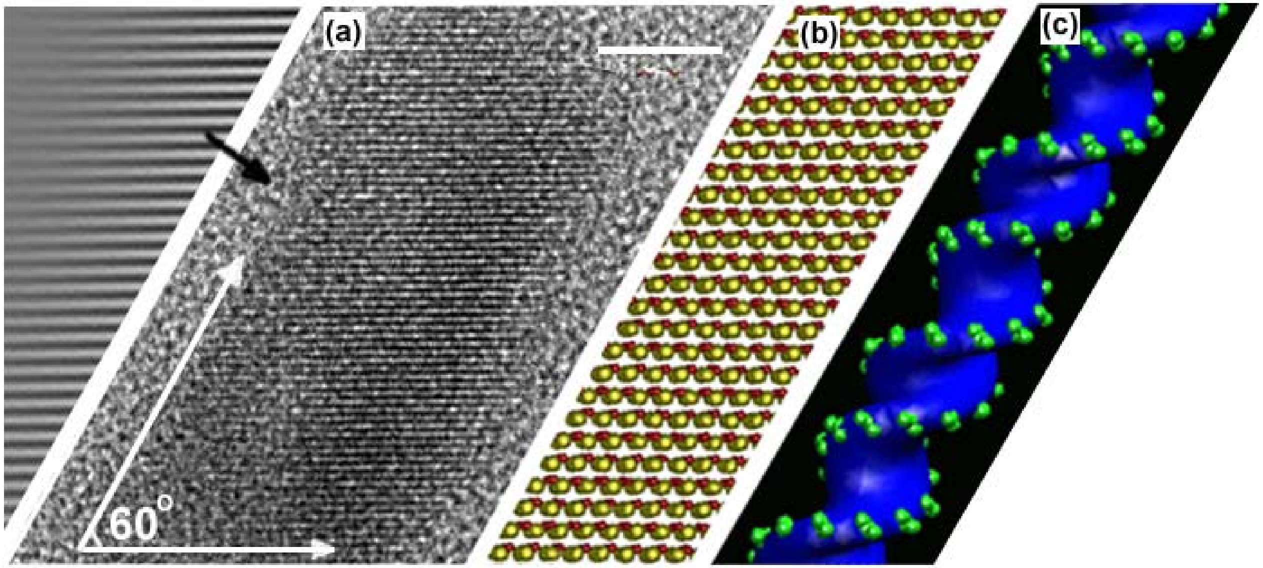


SAXS: expansion of DNA lattice during growth of CDS nanorods



Isoelectric
70/30 DOTAP/DOPC

Molecular casting of CdS nanorods

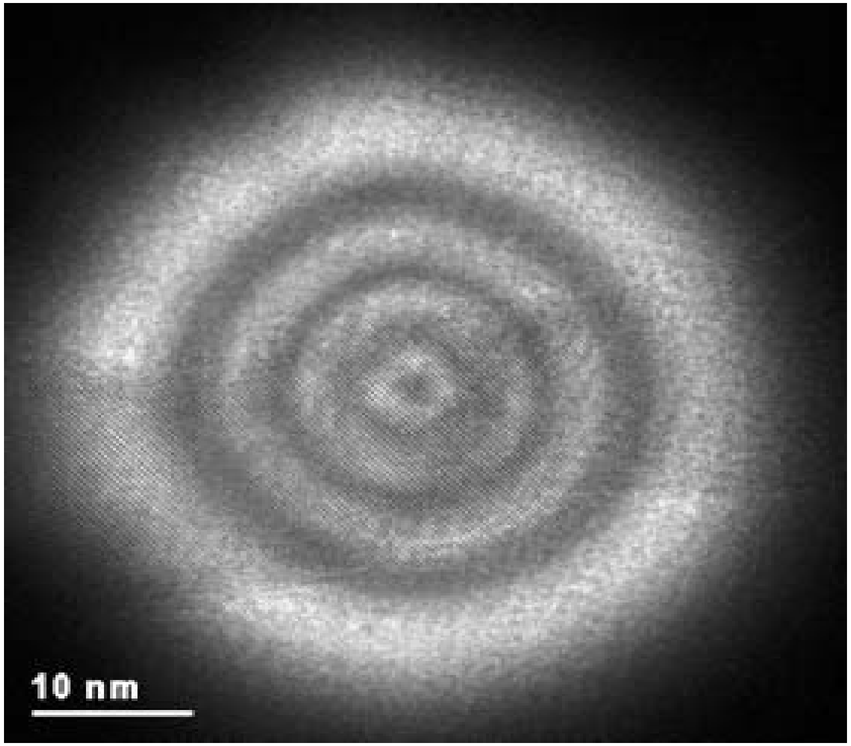


HRTEM CdS lattice DNA

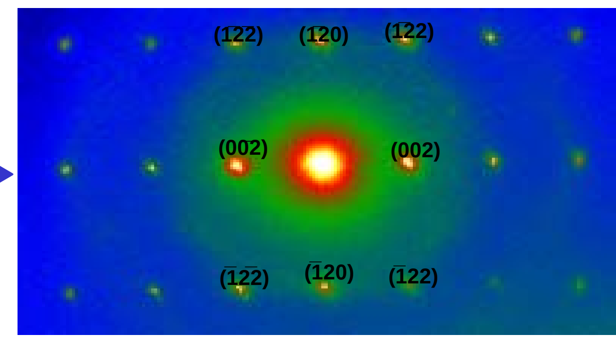
• Strong electrostatic interactions align the templated CdS (002) polar planes parallel to the orientation of the negatively charged sugar-phosphate DNA backbone → molecular details of the DNA molecule are imprinted onto the inorganic crystal structure.

• Crystallographic control via biomolecular architecture: Templated nanorods have (002) directions tilted by 60° with respect to the rod axis, in contradistinction to all known templated CdS nanorods

3-D reconstruction of CdS nanorods at atomic level



FEG EM

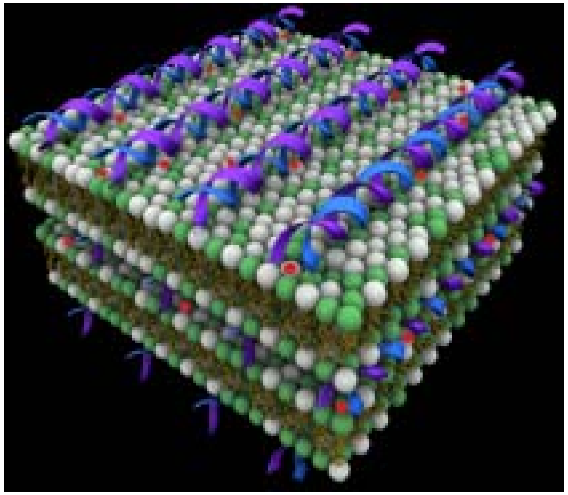


[2,1,0] zone

- Nanodiffraction experiment on single CdS nanorod
- (002) planes tilted by ~60° with respect to the rod axis
- Working on full structure using phase retrieval algorithms

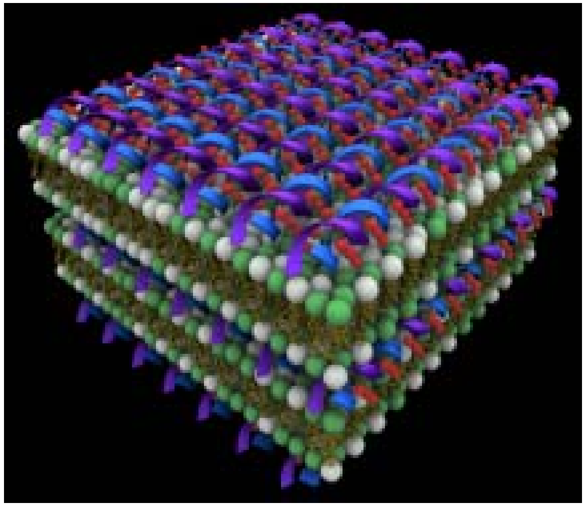
Crystallographic control in biomineralized inorganic nanostructures

$[Cd^{2+}]$=10 mM



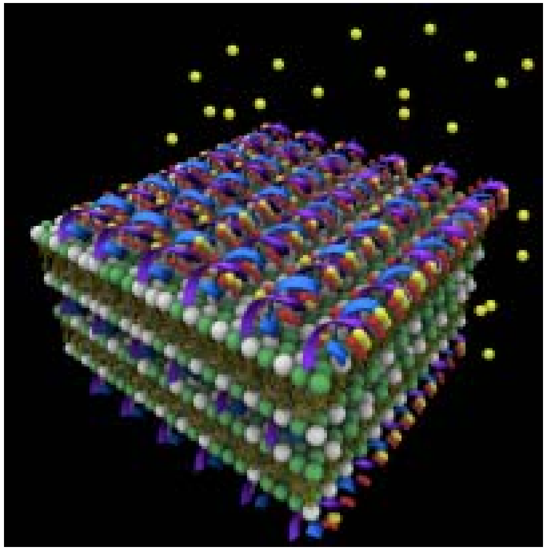
DNA-membrane complex:
Entropically controlled
electrostatic interactions

$[Cd^{2+}]$=40 mM



Organize ions in complex:
Replication of DNA charge pattern

$[Cd^{2+}]$=40 mM
H_2S reaction



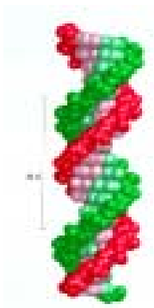
React with H_2S:
Crystallographic control
of CdS nanostructures

Liang et al., JACS, **125**, 11786-11787 (2003)
Liang et al., JACS, in press (2004)

Can we control the pore size and shape within biopolymer-membrane complexes?

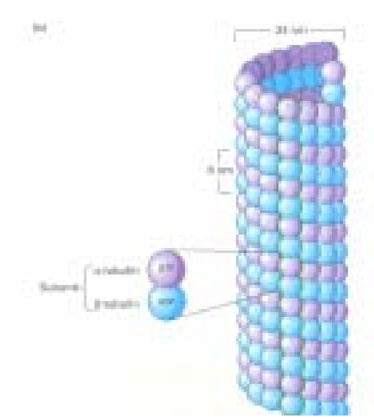
Substitute DNA with bigger biopolymer to make bigger pore?

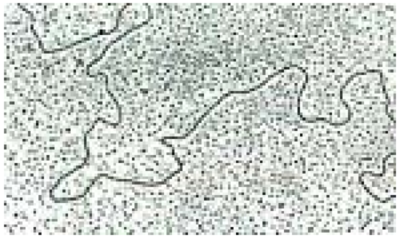
DNA

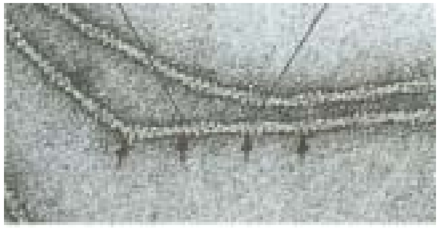


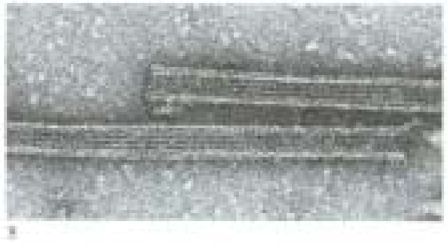
F-Actin

Microtubules



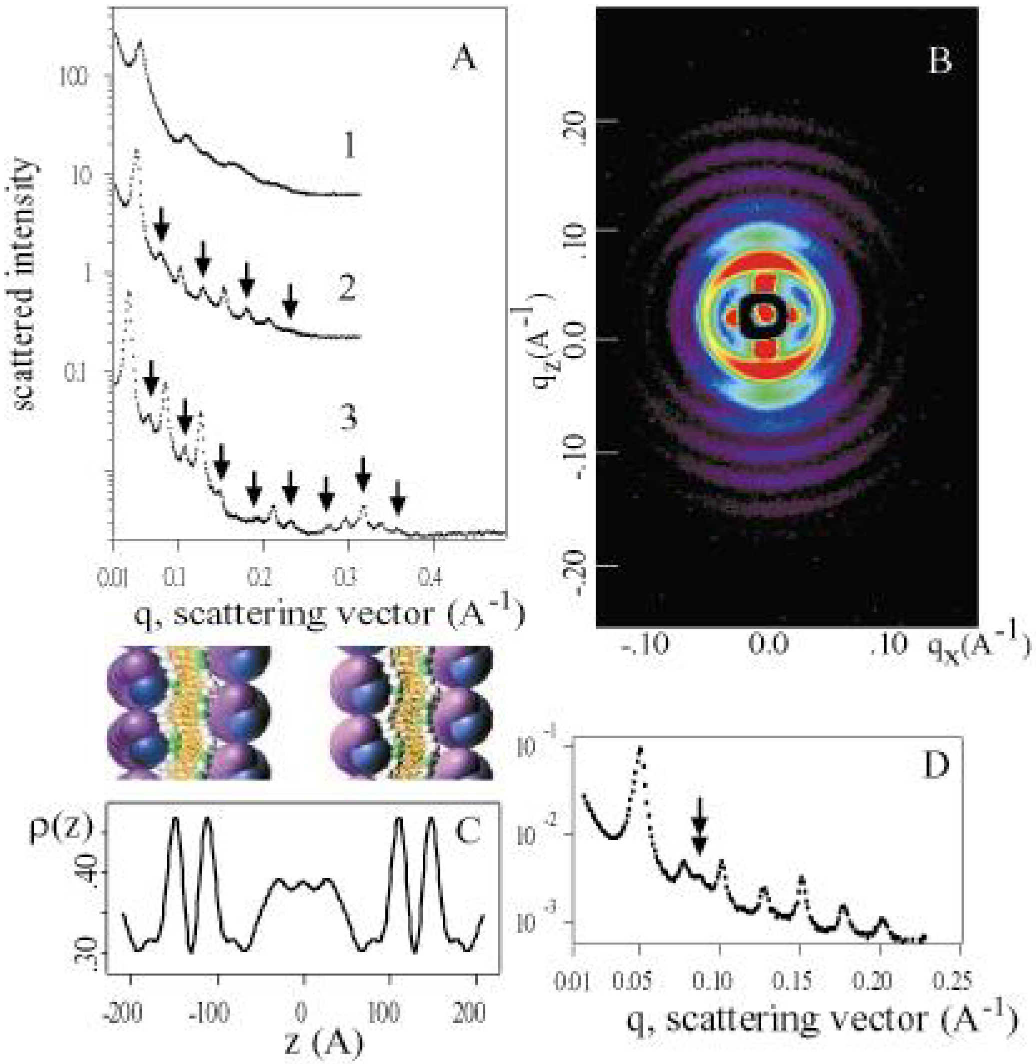






	DNA	F-Actin	Microtubules
ξ_P	50 nm	3-10 μm	mm’s
Diameter	2 nm	8 nm	25 nm
e/nm^2	1/1	1/8	1/5

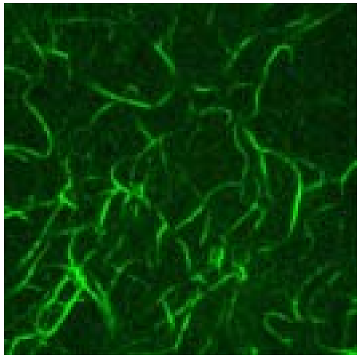
Actin-membrane complexes self-assemble into 'missing-layer' superlattice

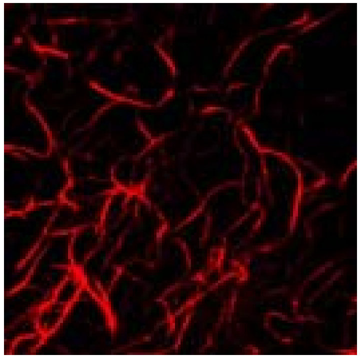


Wong et al., *Science*, **288**, 2035-2039 (2000).

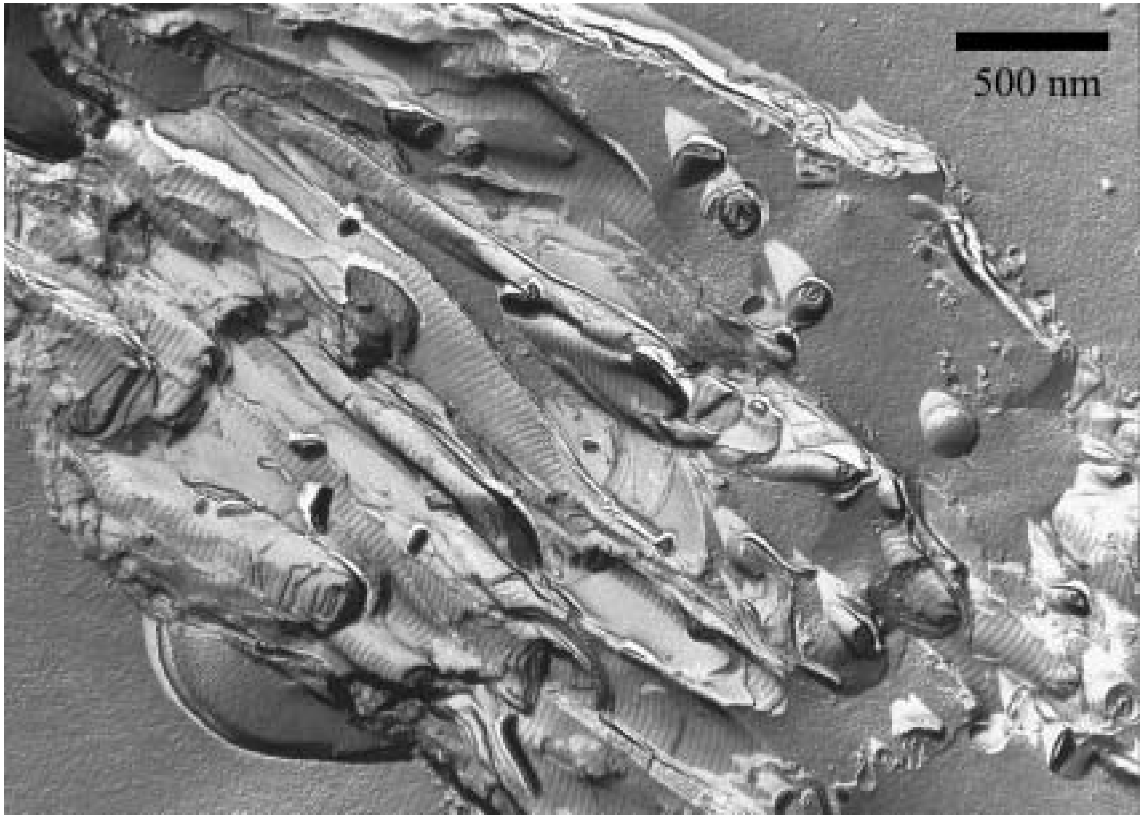
- Simple lamellar structure of DNA-membrane complexes suppressed
- Unit cell contains 2 layers of actin and 1 lipid layer
- Actin is close-packed into 2-D layer on either side of membrane
- No longer have size-tunable nanopores between polymers

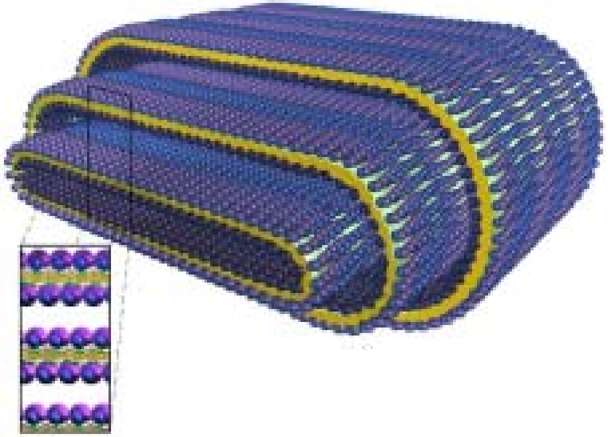
2-D composite actin-lipid membranes collapses into hierarchically structured tubules





DIC and confocal microscopy:
Membrane and actin colocalized.
Alexa 488 Green: actin
Texas Red: membrane

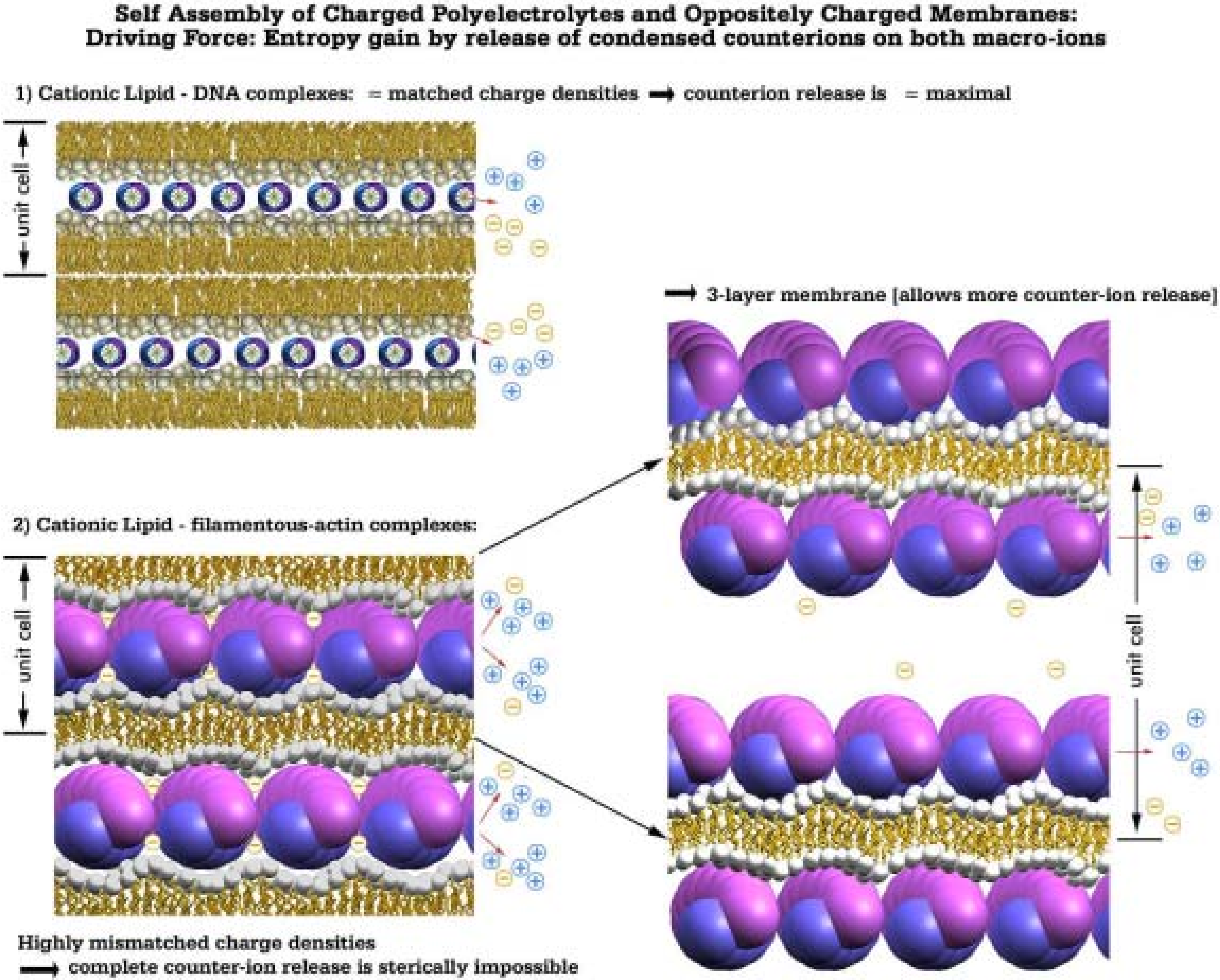




'Freeze-fracture' EM:
2-D phase-locked lattice of F-actin

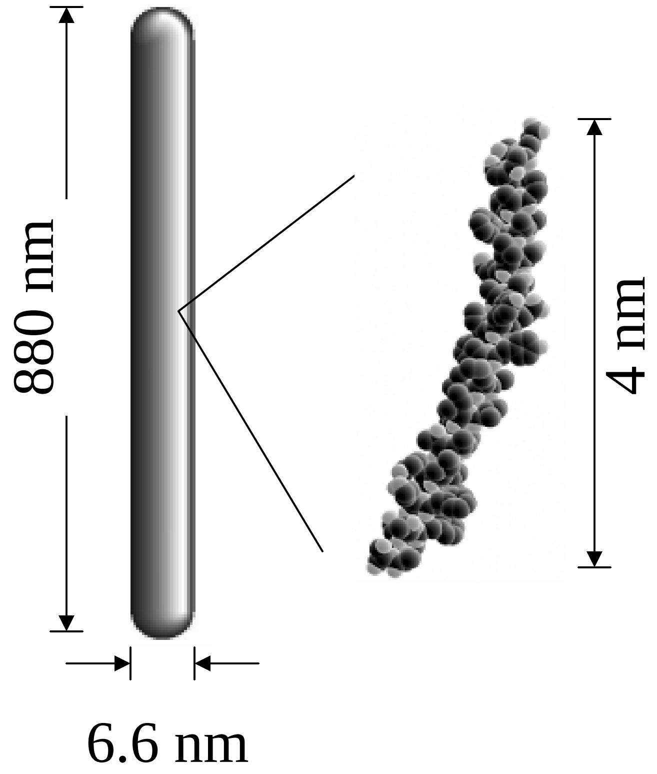
Wong et al., *Science*, **288**, 2035-2039 (2000).

Counterion release and surface charge density matching



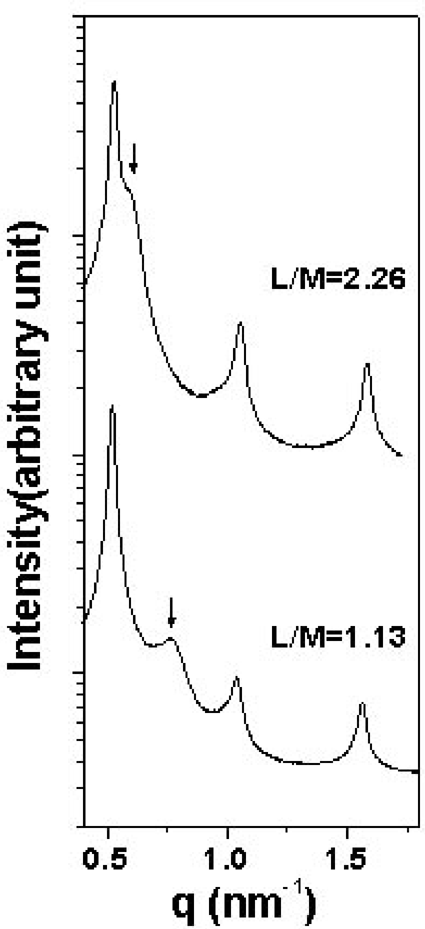
Is it possible to suppress the superlattice structure *and* maintain big pores?

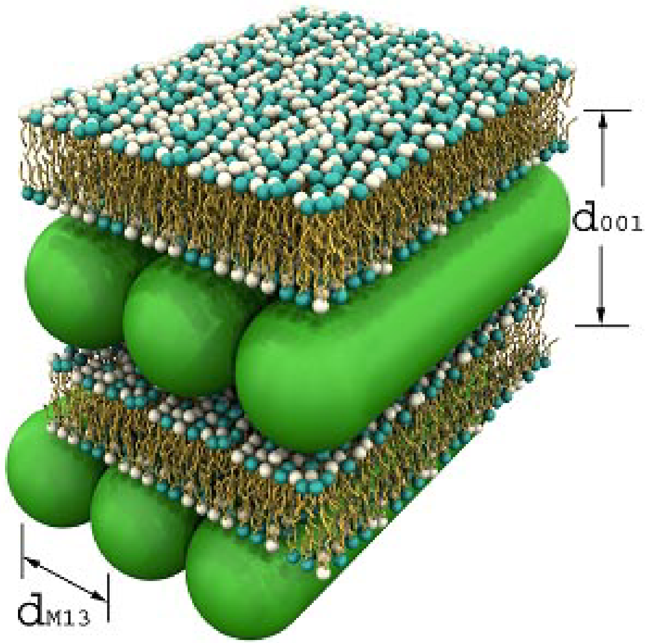
- M13 virus: Monodisperse rod-like particles (D ~ 6.5nm, ξ_P~ 2μm)
- surface charge density can be 'matched' to typical lipid headgroups
- ***Similar charge density to actin but at much higher surface charge density***



- ~2700 copies of α-helical coat protein
- each coat protein ~45 residues, of which 6 are solvent accessible and contribute to surface charge
- surface charge density tunable

M13 virus – cationic membrane complex

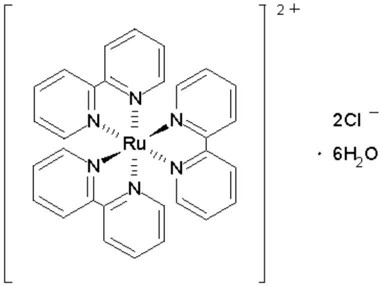




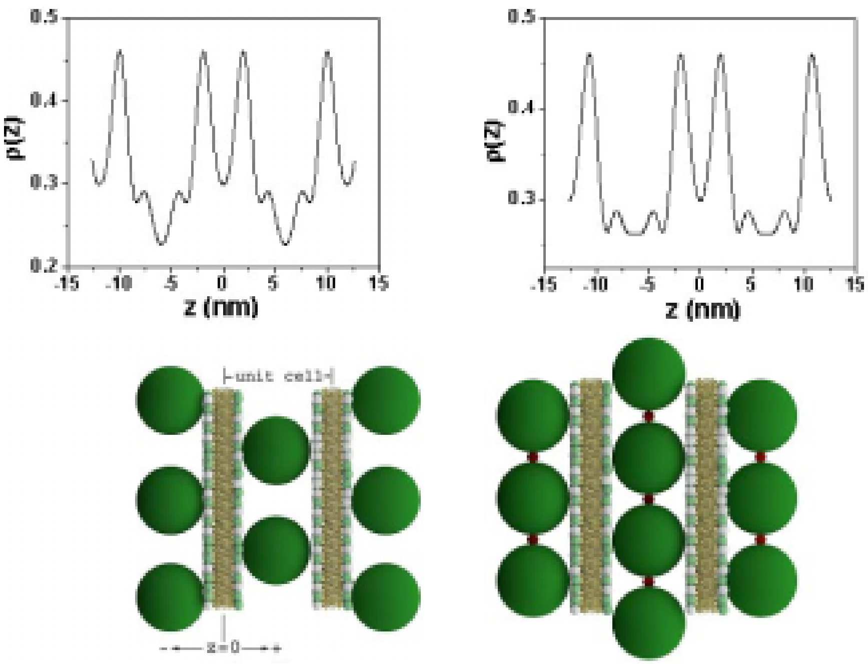
- Simple lamellar structure of DNA-membrane complexes recovered after charge density matching
- Cross-section of nanopore 10x larger than that of DNA-membrane complexes
- Large molecules can be encapsulated

Yang, Liang, Angelini, Butler, Coridan, Tang, & Wong, *Nature materials*, in press (2004).

Organize nanoscopic arrays of dye molecules in complex



$Ru(bpy)_3^{2+}$



• Electron density reconstruction of unit cell from x-ray data

• **$Ru(bpy)_3^{2+}$** organized by M13 virus into correlated, spatially periodic arrays

Yang, Liang, Angelini, Butler, Coridan, Tang, & Wong, *Nature materials*, in press (2004).